THE
AMERICAN NATURALIST

VOL. LXXXII *September–October, 1948* No. 806

AN ALL-FEMALE STRAIN OF LADY BEETLES WITH REVERSIONS TO NORMAL SEX RATIO[1]

DR. A. FRANKLIN SHULL
PROFESSOR OF ZOOLOGY, UNIVERSITY OF MICHIGAN

SOME parthenogenetic species of animals are known only as females. From this extreme the sex ratio ranges, in special situations, through degrees of female preponderance, equality of the sexes, and male preponderance, occasionally up to the limit of exclusively male progenies. Whether such irregularities could be explained has most often depended on whether the animals exhibiting them provide any genetic tools. The genetically best known genus, Drosophila, has naturally offered the best opportunity for such explanations.

We may pass over instances of irregular distribution of chromosomes, leading to intersexes, even when such irregularities are induced by genes, such as the one discovered by Gowen (1928, 1931) in the third chromosome of *Drosophila melanogaster*. These phenomena are too distantly related to the subject matter of this paper to be considered here.

A great preponderance of females (96 per cent) in *Drosophila obscura* was found in one strain by Gershenson (1928). In this strain the Y-chromosome spermatozoa of males possessing a certain sex-linked gene were mostly prevented from fertilizing the eggs. Females possessing the gene, whether homozygous or heterozygous, were not affected. In *D. affinis*, Morgan, Bridges and Sturtevant (1925) briefly reported a strongly female sex ratio which was dependent on the males; any females

[1] Contribution from the Department of Zoology, University of Michigan.

mated with males of this strain produced the one-sided families. Sturtevant (1940) later located the gene responsible for such a change in the right limb of the X chromosome, and showed that it was accompanied by an inversion. Whether the inversion was always accompanied by the abnormal sex ratio was not determined. Sturtevant and Dobzhansky (1936) demonstrated a similar situation in *D. pseudoobscura* and *D. persimilis* (then regarded as races A and B of *pseudoobscura*). The gene is in the right limb of the X chromosome; it causes the X chromosome to undergo equation in both maturation divisions, while the Y degenerates. Most spermatozoa are thus X-bearing. A similar gene is reported in two other species besides the ones mentioned here.

Dr. A. Buzzati-Traverso kindly permits reference here to an all-female strain so far reported only in *Drosophila Information Service* (No. 14, 1941, p. 49). The species was then regarded as *D. bilineata,* but has since been described as a new species, *D. bifasciata,* one of the so-called *obscura* group. The females of this strain, crossed to males from other strains, yielded only female offspring. The war prevented continuation of the study of this strain, and nothing yet discovered provides an explanation of its peculiarity. (Personal communication.)

All-male progenies in a strain of *D. melanogaster* were attributed by Gowen and Nelson (1942) to a dominant gene in the female. Novitski (1947) describes a similar preponderance of males in *D. affinis,* which arose by a modification of the strain previously giving preponderance of females. The change was produced by a recessive autosomal gene in the chromosome pair designated B. The abnormality is a property of the males, which have the old (female) sex-ratio gene (*sr*) in their X chromosome and are homozygous for the new autosomal mutation.

Observation on Lady Beetles

The peculiarity here reported in lady beetles most nearly resembles the one discovered in Drosophila by Dr.

Buzzati-Traverso. From a small group of hibernating beetles of the species *Hippodamia quinquesignata* kindly sent from Logan, Utah, by Professor G. F. Knowlton, one pair found copulating was isolated. The progeny of the female, which could have mated with other males, consisted of 39 females and one male. This one male is known to have been the first to pupate, because dates of pupation were marked on the vials in which the larvae were isolated. Hence, whatever event resulted in exclusively female progeny occurred at the beginning of the family; the remainder of the offspring were presumably produced under its influence.

The lone male in this family lived only three days, hence his sisters were necessarily mated to males from another source if the one-sided sex ratio was to be studied at all. Another pair of beetles from the same Logan group was producing the normal numbers of both sexes, and males from this family were used in half a dozen matings with the daughters of the first pair. Five of these pairs produced only female offspring in families ranging in size from 3 to 21 members. A sixth pair produced 7 females and one male.

The above matings, and all others subsequently made which involved the abnormal sex ratio, are shown in the chart in Figure 1. It should be remembered that these beetles were being reared primarily to study the genetics of color pattern. Also, it should be recalled that many matings of lady beetles do not succeed, and that this difficulty increases as the generations pass. The matings here recorded are not always, therefore, the most desirable ones; they are the ones which succeeded.

The heavy numbers in the chart are the experiment numbers. The color-pattern results have already been published (Shull, 1945), and some information about these experiments can be obtained by checking the experiment numbers in Table I of the earlier paper. Beneath the experiment designation, in lighter numbers, is the sex ratio of the progeny, the females being given first.

The numbers of individuals reported now are in some

experiments smaller than in the former paper because some beetles whose color patterns could be observed were damaged so that sex could not be ascertained. In two instances the number now reported is larger than in the earlier paper because certain individuals used as parents had been lost or were pinned in a separate collection and at first overlooked.

The six outbred families which have already been described are those of Experiments 280 to 290 inclusive. The complete line sloping upward to the left from this group of families indicates that the mothers of all of them were among the 39 daughters of 259. The short line sloping upward to the right indicates the males from an outside source, which for these six families was Experiment 260. To simplify the chart the sources of the outside males are not indicated, but they may be ascertained from Table I of the earlier paper (Shull, 1945).

The further course of the experiments which succeeded is clearly shown in the chart. The divided family of 289 was utilized for a mating, from which a small divided family (302) was obtained. A daughter and the son of 302 were paired, but escaped before eggs were laid. The other daughter was mated to an outside male, and by them a divided family (316) was produced. Most of the other families were exclusively female, exceptions being 309, 311 and 313.

In 319 there were more males than females, though this excess may well be accidental. The daughters used, which could have been mated with their brothers but for a study of color pattern were actually crossed with outside males, gave perhaps normal sex ratios (Expts. 322, 323, 325).

In 320 there was one male, but some of the 17 females retained the abnormal sex-ratio peculiarity, for Expts. 326 and 327 still yielded only females. From 326 there descended ten families, in two further generations, consisting only of females. From 327, the next two generations included 6 all-female families, and one (352) divided 6 : 1. The one male in 352 was mated with one of the females

of 348, and their progeny included 23 females and 35 males. Further generations derived from 358 all had normal sex ratio. Other matings made with females from exclusively female families of the same generation as 348 failed; and when it was discovered that 358 showed normal ratio, it was too late to make still other matings. Consequently, the all-female strain was lost at this point.

Eggs for cytological study were fixed, but this step was taken too late. It was impossible to remove eggs laid on the confining lantern globes without injuring them. They needed to be laid on the leaves or other parts of the potato plants, which could be removed and fixed with the eggs. It was desirable, also, to obtain large batches of eggs so that many could be sectioned in one block. Experiment 358 yielded the first eggs, in several generations, which met the above requirements, and some of these were fixed. When the adults hatching from eggs of this family proved to be both males and females, the fixed eggs were regarded as worthless for a study of the cause of the abnormal sex ratio. It was too late, thereafter, to get any other eggs for fixation.

Abnormality a Property of Female

In explaining a one-sided sex ratio, it is important to know which sex is responsible for it. In related instances reported by other investigators, as described in the introduction, it has been sometimes the male, sometimes the female, which caused the aberration.

In the strain reported here, the females were mated with males of the same strain only twice (Expts. 302 and 358). In these two families the resulting sex ratio approached normal, and if there had been any peculiarity calling for explanation it could have been attributed to either parent.

In the other matings, in which the males came from other strains, four outside sources were used. The five original families in which the phenomenon was first observed (Expts. 280–287, and 290) were fathered by males from a normal strain from the same hibernating mass

as the all-female strain, namely, that from Logan, Utah. The males in Expts. 295, 305, 312, 314 and 315 were hybrid descendants of a species cross of a female *H. quinquesignata* from Logan, Utah, and a male *H. convergens* from Placerville, California. In Expts. 307, 326, and 327 the males were from Yosemite National Park, California. Experiments 333–351 and 353 were offspring of males from Boulder, Colorado, or the laboratory-bred descendants of beetles from that area. In the above statement those experiments which gave divided progenies have been omitted. Hence, four widely separated collections have furnished the males used. It is highly unlikely that the males could be the source of the one-sided sex ratio, and turn up in all of these random collections as the randomly selected individuals to be mated. Moreover, males from these same sources were used in other matings which resulted in offspring of both sexes. It is concluded, therefore, that the cause of the abnormal sex ratio is some feature of the females exclusively.

Parthenogenesis Not the Cause

Since it is the female which is responsible for the abnormality, the cause of the exclusively female progenies could be parthenogenesis. Eggs which result from a single maturation division, which are diploid, and which develop only into females, are regularly produced in many kinds of animals, chiefly among the insects. Were this the explanation of the all-female progenies here reported, it might be assumed that the spermatozoon stimulated cleavage but did not contribute chromosomes and genes to the progeny.

No instance of parthenogenesis in the Coccinellidae appears ever to have been reported. Moreover, in previous work on the genetics of this group hundreds of virgin females have laid many thousands of eggs, not one of which developed. Even in this abnormal strain, many females were kept virgin and laid numbers of eggs, but none of these developed. This, however, is negative evidence, and there conceivably still might be parthenogenesis.

Fortunately, there is genetic evidence which bears on this question. Among the outside males used in these experiments, seven exhibited the dominant spotless pattern (Shull, 1943, 1944, 1945). These are indicated in Figure 1 by the letter *S* above the shortened oblique lines. Five

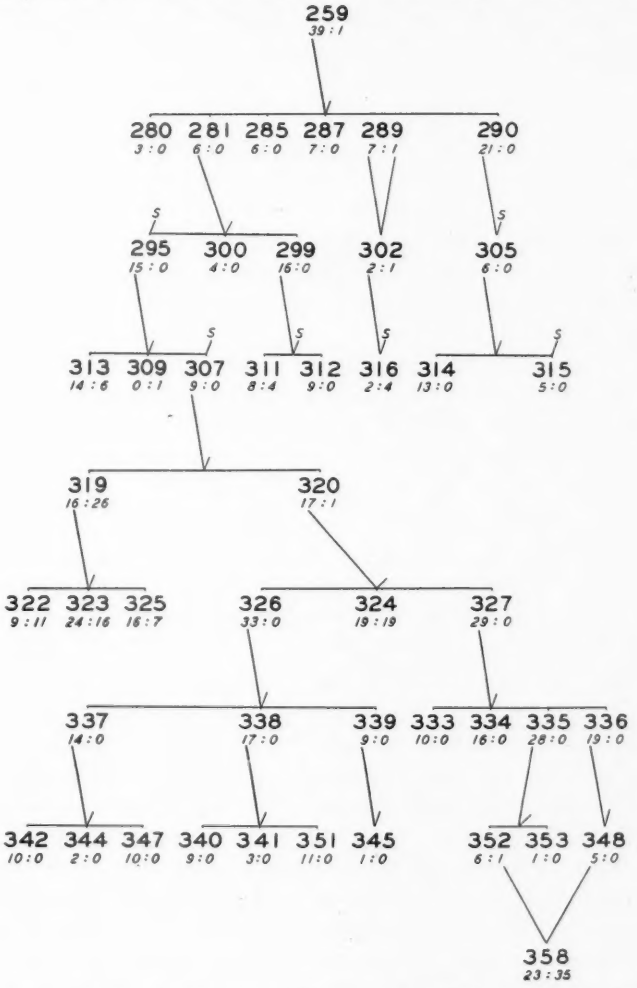

FIG. 1.—Course of experiments. Heavy numbers, designations of experiments. Light numbers, sex ratios, females named first. Sloping lines, sources of parents. *S*, spotless fathers used.

of the families produced by them consisted only of females. These are Expts. 295, 305, 307, 312 and 315. The offspring of each of these was partly spotless, partly spotted. In the aggregate, they included 23 spotless and 25 spotted. Obviously the males were all heterozygous for spotless, as was known for three of them from their parentage (Shull, 1945, Table I).

In the progenies including both sexes, and fathered by spotless males (Expts. 311 and 316), there were 10 spotless and 8 spotted. Spotless is handed on to all-female progenies in exactly the same way as in those with normal sex ratio.

It is clear that the eggs were fertilized in these seven families, so it may be assumed that parthenogenesis is not the cause of the one-sided sex ratio.

Other Conceivable Causes

Further consideration of the factor responsible for exclusive production of females must be speculative. Among the explanations offered by other authors for one-sided sex ratios, the disabling of one of the kinds of spermatozoa (the Y-bearing in this instance) can hardly be responsible, since the peculiarity belongs to the females; they could scarcely influence the production of spermatozoa. Nor can the ratio depend merely on a lethal gene in the X chromosome, for the males would thus be merely reduced in number, not excluded completely.

Sharply defined and temporary differential mortality might be the explanation. Death losses are fairly large after a line has been reared a few generations. Occasionally, even the early generations show high mortality; Expt. 300 was an example of this. It was a particularly unhealthy line; but from the large number of larvae and pupae lost in it, and the small number surviving (only four), both sexes would have had to be subject to the mortality if both were present. In any case, any differential mortality eliminating the males would have to be a disability from which certain families could completely

recover. No weakness of males in general would be an adequate explanation.

It should be pointed out that many of the families described are species hybrids of one grade or another, as was indicated earlier in describing the sources of the males used. Such hybridity, however, can hardly explain the all-female progenies, since the one-sided sex ratio appeared before the species hybrids were introduced.

Selective fertilization favoring the X-bearing spermatozoa could be the explanation. So, also, could attached-X chromosomes be assumed, if some way of eliminating the expected Y-bearing eggs were included in the scheme. If the existence of a Y chromosome without an X in an egg were lethal, or even if it merely prevented fertilization, all-female families would result. This would be essentially a gamete lethal, which is rare in animals though common enough in plants where such lethals destroy the gametophyte.

The suddenness with which the all-female influence disappeared from some branches of the strain described should influence one's idea of the cause of the phenomenon. It is obvious that the return of normal sex ratio in some families was a sharply defined result. Even those families which included a single male scarcely indicate that the reversal was a gradual process; for clearly in one of these families (Expt. 320) some of the sisters of the lone male retained the all-female influence in its entirety. The reversal was presumably sudden, but did not occur in all of the germ cells.

Because the reversion to normal ratio is sudden, the all-female influence is probably not a cytoplasmic one. Something is needed which can happen quickly. Nondisjunction resulting from attachment of the X chromosomes meets this requirement, since in other organisms with attached X's the separation of these chromosomes is not uncommon. This explanation requires, however, as stated earlier, the less likely subsidiary hypothesis that the Y-bearing eggs be eliminated.

Among these possible explanations it is difficult to name a most probable one. They all have their inadequacies. If there were any evidence elsewhere of a harmful effect of the Y chromosome in eggs, the attached-X explanation would seem best.

Lack of Males an Advantage?

It is perhaps justifiable to speculate on the possible advantage of absence of males in a given female line of descent. It forces repeated outcrossing. In the lady beetles, where genetic experiments have often been cut short by the declining vigor of successive generations, outcrossing could be regarded as helpful. The series of experiments which included this all-female strain ran through nine generations—more than in any other series conducted by the present author. This comparison need not mean much, since experiments are discontinued for various reasons; but it is suggestive.

If lack of males in some strains in advantageous, and its mechanism is not a rare phenomenon, other such strains probably exist. None has been observed in the experiments with Michigan coccinellids, though no systematic search for them has been made. In a group of eight beetles collected at the end of the active season of 1947 by Professor Knowlton in Utah, all were females belonging to two species. One of these species produced a dozen offspring, all females. These were confined with males of another (Michigan) species, but the species cross failed, and the Utah line was lost. The numbers of individuals are too small to prove an all-female strain. It would seem worth while, however, to look specifically for the one-sided sex ratio in the western species which gave these suggestive results.

Summary

One strain of beetles produced exclusively females through eight generations. At various points in this line of descent certain branches reverted to normal sex ratio. The peculiarity was a property of the females,

since the males to which they were mated came from four wild sources, and since males from the same families, mated to females of other strains, produced both sexes. Parthenogenesis is excluded as the cause of the one-sided sex ratio because some of the males introduced from outside strains possessed the dominant spotless gene, and every family of females produced after such an introduction included some spotless individuals. Some of the males used were species hybrids or descendants thereof, but the all-female feature arose before such hybridity was introduced. Explanations involving the disabling, or elimination, of the Y-bearing spermatozoa are unlikely, since the male would presumably have to be responsible for such effects. A cytoplasmic influence seems unlikely, because its loss probably could not be as sudden as were the reversions to normal sex ratio. Selective fertilization, favoring the X spermatozoa, could explain the phenomenon, but the reason for the selectivity would have to be something that could disappear suddenly. Attachment of the X chromosomes could be the reason, if something prevented the functioning of Y-bearing eggs. Separation of the X's would be sudden enough to account for the sharply defined return of normal sex ratios.

Existence of all-female strains may have selective advantage, since it compels outcrossing. This would be especially plausible if the decline of vigor so often reported in experiments with lady beetles is a result of inbreeding.

LITERATURE CITED

GERSHENSON, S., 1928. *Genetics*, 13: 488–507.
GOWEN, J. W., 1928. *Science*, 68: 211–212.
GOWEN, J. W., 1931. AM. NAT., 65: 193–213.
GOWEN, J. W., and R. H. NELSON, 1942. *Science*, 96: 558–559.
MORGAN, T. H., C. B. BRIDGES, and A. H. STURTEVANT, 1925. *Bibliographia Genetica*, II: 1–262.
NOVITSKI, E., 1947. *Genetics*, 32: 526–534.
SHULL, A. F., 1943. *Jour. Hered.*, 34: 329–337.
SHULL, A. F., 1944. *Jour. Hered.*, 35: 329–339.
SHULL, A. F., 1945. *Jour. Hered.*, 36: 149–160.
STURTEVANT, A. H., 1940. *Genetics*, 25: 337–353.
STURTEVANT, A. H., and TH. DOBZHANSKY, 1936. *Genetics*, 21: 473–490.

THE ROLE OF SYMBIONTS AND AUTOCATALYSTS IN THE GENETICS OF THE CILIATES

DR. EDGAR ALTENBURG

THE RICE INSTITUTE

ONE of the things that distinguish Paramecium from the general run of cells is its enormous size. In fact Paramecium is large enough to be easily visible against a dark background with the naked eye. This enormous size is of advantage to Paramecium in the capturing of its food and in protecting it against attack by smaller organisms. But it was also of advantage, in the course of evolution, for Paramecium to retain the rapid rate of reproduction and dispersal made possible by a unicellular organization. The Protozoa in general are larger than the average cell for the same reason.

There are certain peculiar features in the heredity and development of Paramecium connected with its enormous size. In the first place, the large cell size has led to the evolution of a correspondingly large nucleus. For in all cells there is a definite size relationship between nucleus and cytoplasm. Increase in cell size necessitates a corresponding increase in nuclear size; the reverse is also true as in polyploids and in the salivary gland cells of Drosophila. In Paramecium, increase in nuclear size came about by the formation of the macronucleus, a compound body consisting of about a hundred diploid nuclei each still enclosed in its own membrane. At cell division, it is probable that each diploid nucleus divides mitotically (forming about two hundred diploid nuclei, one hundred for each new Paramecium if this was the original number). The macronucleus as a whole then divides by "amitosis," though the individual nuclei do not. (In Protozoa in general the nuclear membrane does not disappear at mitosis; this probably applies to the membranes of the individual diploid nuclei in the macronucleus of Paramecium.)

By virtue of its size, the macronucleus would be expected to control the physiological processes of the cell, to the exclusion of the micronucleus; for at each locus it would contain a hundred pairs of genes (if the number of its diploid nuclei is one hundred), and these hundred pairs would be dominant to the single pair in the micronucleus of *P. caudatum* or even to the two pairs in the double micronucleus of *P. aurelia.* This would be true even if the genes in the macronucleus were individually recessive and at least one of those in the micronucleus dominant (a situation which is experimentally possible by regeneration of the old macronucleus pure for a recessive gene in an ex-conjugant heterozygous for the dominant allele received from the other parent). But because of its highly compound nature the macronucleus cannot very well undergo the orderly process of meiosis and Mendelian segregation, and so must undergo degeneration at meiosis if sexual reproduction is to be effective. Hence an ordinary diploid nucleus—one capable of meiosis—becomes necessary in addition and is represented by the micronucleus.

But the two kinds of nuclei (macro- and micro-) almost certainly do not differ in the fundamental character of their individual genes—the one does not produce plasmagenes if the other does not. Neither does the macronucleus differ fundamentally from the ordinary nuclei of other organisms, except for its compound nature and resultant larger size—the thing that makes it the physiological (or somatic) nucleus of Paramecium—and this we saw was merely a result of increased cell size. It is true that the macronucleus degenerates at the time of sexual reproduction, but this is somewhat comparable to the degeneration of the nucleus in the case of the red blood corpuscles, made necessary on physiological grounds. Perhaps it comes about in the case of Paramecium through the action of a nuclease with a very localized distribution. In any event, the new genotype produced by conjugation could not express itself unless a

new macronucleus replaced the old one.

The large cell size of Paramecium led to a second complication. Size made possible a degree of differentiation not found in single-celled organisms of smaller size. Thus the cytoplasm came to serve as the equivalent of the soma of multicellular organisms. But it also had to serve as material for the germ cells at the time of conjugation. Hence it became necessary that the cytoplasm of Paramecium return at the beginning of each sexual cycle to the undifferentiated state usual for germ cells. The multicellular organisms are not confronted with a problem of this sort, since the germ track remains separate from the soma and does not undergo differentiation. A mechanism (perhaps somewhat special for the Protozoa) must have developed in the course of evolution for bringing about the de-differentiation of the cytoplasm at the time of conjugation in Paramecium. However, a line maintains itself in its differentiated state in the course of asexual reproduction. It must therefore contain some sort of cytoplasmic material which can reproduce and multiply in the course of asexual reproduction, and which serves as the physical basis for the inheritance of differentiation. But since there is de-differentiation of the cytoplasm (at least with regard to many traits) with each sexual cycle, it is evident that the self-reproducing material above referred to must disappear with de-differentiation, and that it must form again *de novo* at the time of re-differentiation. Such substances therefore are not genes; they are autocatalytic in that they produce more material like themselves in maintaining differentiation during asexual reproduction—a matter to be considered more fully later. Muller (1929) has pointed out the distinction between genes and ordinary autocatalysts.

It was probably also because of cell size that the ciliate relatives of Paramecium often came to harbor green algae (Zoochlorellae) as symbionts: only a relatively large cell could furnish quarters sufficiently spacious

for intracellular symbionts as large as Zoochlorellae. This fact might account for the widespread occurrence of the green symbionts in the larger Protozoa in general. Green symbionts are found, for example, in *Ameba viridis,* in many of the Heliozoa and Foraminifera, in almost all the Radiolaria (most of which contain the yellow-green alga Zooxanthella), and in many flagellates. They are especially frequent among the ciliates, among which we might note, as familiar examples, *Vorticella viridis* and *P. bursaria,* both of which are very large.

Now in the course of evolution symbionts might very well have come to interact with the genetic system of their hosts. In particular, it has been suggested that kappa (the killer factor) in *P. aurelia* is a symbiont, related to the green symbionts (Zoochlorellae) found in *P. bursaria* (Altenburg, 1946). This suggestion was based originally on the correspondence in number of green symbionts in *P. bursaria* and the number of kappa bodies demonstrated by Preer (1946, 1948*b*) in *P. aurelia* (about 1200 per cell in each case). But more facts have accumulated in support of the suggestion, two of which are of particular importance. One of these was the discovery that the kappa bodies were large enough to be plainly visible under the microscope (Preer, 1948*a*). This at once ruled out the theory that kappa is a plasmagene derived by fractionation from a nuclear gene; all estimates agree in putting the size of the gene beyond the limits of visibility under the microscope. It is true that only Paramecia of genotype *K* can harbor kappa. But this would not necessarily mean that kappa was modified *K*. It might well be that in Paramecia of genotype other than *K* cytoplasmic conditions are not favorable to the continued growth of kappa.

The relatively large size of kappa is consistent with the symbiont theory, since kappa might well be visible if it was derived from a symbiont which itself was visible. It is true that the kappa bodies are not quite as large as the Zoochlorellae of *P. bursaria,* but it is entirely

possible that they might have become reduced in size since the time of their supposed origin from Zoochlorellae, perhaps as the result of the close symbiotic relationship with their host, involving among other things the loss of chlorophyll in this particular case. In fact, chloroplasts become very small after losing their chlorophyll; they might even totally disappear. Moreover, material stained with a nuclear dye would show only the nucleus of the supposed symbionts, not the cell as a whole, and the nucleus might be very small.

A second important fact that came to the support of the symbiont theory was the discovery, again by Preer, that the division rates of the kappa bodies and of Paramecium are somewhat independent of one another (Preer, 1948*a*). At higher temperatures the division rate of Paramecium is speeded up to a greater degree than is that of the kappa bodies. It is therefore possible (by means of temperature) to reduce the numbers of the more slowly reproducing kappa bodies to considerably less than 1200 per Paramecium and even to eliminate them completely in some of the Paramecia. A very striking fact in parallel is that Zoochlorellae can also be removed from *P. bursaria* by a series of rapid fissions (Jennings, 1938).

The relative independence of kappa and Paramecium is shown by two other facts, in addition to their independent division rates: (1) the selective lethal action of high temperatures on kappa, a temperature of 38° C resulting in the death of kappa but not of the host cell, and (2) the transmissibility of kappa to Paramecia previously without kappa by exposing them to broken-up Paramecia that contain kappa. All these facts point to a relative degree of independence that might be expected of an organism distinct in point of origin and physiological workings from its host, but hardly to be expected of a plasmagene or other body that is an integral part of the genetic system of the host proper.

There are equally strong reasons for ruling out the

theory that kappa is a virus or some other pathogenic organism. The relatively large size of the kappa bodies argues against their being a virus, as usually understood. Moreover, kappa is non-pathogenic and thus unlike any known virus. On grounds of non-pathogenicity also, kappa could hardly be rickettsia. A point to be emphasized here is the rather definite upper limit placed on the number of kappa bodies per cell (about 1200). This limitation in numbers is precisely what might be expected; for a symbiont, in contrast to a parasite, multiplies only to the extent that its numbers are in keeping with the welfare of its host.

If kappa is a symbiont, then up to this point we have no evidence for plasmagenes or for cytoplasmic inheritance in the genetic system of Paramecium proper. However, there is a form of cytoplasmic inheritance in Paramecium itself. This is illustrated by the inheritance of mating type in those stocks of Paramecium which contain the "two-type" mating strains. In these stocks, a given ex-conjugant upon division gives rise to two lines both of which might be plus, or both minus, or one plus and the other minus. These possible combinations of mating types occur purely according to chance and without reference to the mating type of the ex-conjugant itself.

Now, when an ex-conjugant gives rise to two lines of the same mating type (both plus or both minus), there obviously is no segregation of genic material. It is therefore not probable that there is any such segregation when the two lines happen to be of opposite mating type; if there were, then one would expect a given ex-conjugant always to give rise to two lines of opposite type. Obviously mating type is not being determined by any such mechanism. But if there is not segregation of nuclear material, it is highly probable that the macronuclei, as well as the micronuclei, are of identical genotype in the two lines derived from a given ex-conjugant, and that both types of nuclei are derived from the fertilization

nucleus by simple mitosis. Moreover, since the mating type of a new line is often the opposite of what it was before conjugation, it follows that the mating type does not persist through conjugation—the cytoplasm becomes de-differentiated with respect to mating type sometime after the onset of conjugation. Hence the determination of mating types in the stocks in question is not dependent on kappa-type bodies of two specific kinds (one for the plus and another for the minus type); for if it were dependent on such kappa-type bodies, then (contrary to fact) mating type would as a rule persist through conjugation. For the same reason, it could not be dependent on any other type of cytoplasmic body that persisted through conjugation as a specific mating type determiner.

If then mating type of the stocks in question is determined neither by the segregation of nuclear bodies (sex chromosomes or genes), nor by cytoplasmic bodies that persist through sexual reproduction and that are specific for plus or for minus mating type, then there is only one conclusion: mating type must be determined in the two-type strains in Paramecium by the local environment in some sense or another (chemical or physical), and we should have here merely another case of a given genotype with different phenotypic expressions comparable somewhat to the case of sex determination in Bonellia. In the case of Paramecium, mating type is ordinarily determined at the metaphase stage in the division of the ex-conjugant into two (the cytoplasm having previously become de-differentiated). At metaphase the ex-conjugant is constricted into two (transversely), so that it now contains two "fields" of cytoplasm, each with its own macronucleus and each the equivalent of a new cell. The local environment within each field might very well cause differentiation of the cytoplasm with respect to mating type. It might seem that this implies a very narrow restriction (within the microscopic limits of the ex-conjugant) of environmental differences. But

it is precisely such localized differences which cause differentiation of indeterminate eggs (and probably of determinate, also) in the case of multicellular organisms. In Paramecium, a slight difference between the two presumptive division products of the ex-conjugant might very well throw the balance in favor of one mating type or the other.

It is true, however, that each mating type as a rule breeds true so long as it reproduces asexually; that is to say, it remains differentiated as a plus or a minus line. Therefore the differentiated state must have some hereditary basis, since it persists from one cell division to the next. But nuclear genes can hardly be at the basis of inheritance in this case. For we saw that the macronuclei of the two sister lines derived from an ex-conjugant are probably identical genetically, having been derived from the same fertilization nucleus by mitosis (without segregation), and so it follows that any difference between the two mating types must reside in the cytoplasm. It must therefore depend on some cytoplasmic material that is characteristic of each type. Moreover, a single Paramecium can give rise, for example, to over a thousand offspring after just ten cell divisions, and unless the material in question had multiplied it would have become correspondingly dilute. This cytoplasmic material must therefore be capable of increase, but it must somehow cause its own increase, since it is the only material that makes the one mating type different from the other (the two types being alike as regards their genes). The material in question is therefore some sort of *autocatalyst*. But we saw that mating type disappears during sexual reproduction and is then formed anew; the autocatalyst in question would most likely follow a similar course. Now genes proper do not go out of existence at the time of sexual reproduction. Neither are they formed *de novo* with each sexual cycle. Hence the autocatalyst is not genic and can hardly be referred to as a "plasmagene." Since it disappears with

each sexual cycle, it must ultimately depend on nuclear genes in conjunction with the specific local environment for its initiation. In fact, the dependence of the autocatalyst on a nuclear gene is known: the two-type mating strains cannot develop in the absence of a certain nuclear gene (+, allelic to mtI for the one-type strains), as shown in the F_2 from crosses between the two-type and the one-type mating strains.

The autocatalyst above referred to is not in any way to be confused with the kappa substance, for kappa does not reappear in a line once it has disappeared, unless new kappa is introduced from some other Paramecium (through delayed conjugation). Just the opposite sort of thing applies to the autocatalyst.

However, in the *B* group of *P. aurelia,* mating type is seemingly dependent on kappa-like bodies insofar as it does actually persist through conjugation in the *B* group. But it is possible that even here some sort of autocatalyst of a more persistent type, yet not genic in nature, is involved. It would be necessary to know in this connection whether the apparently permanent mating types are really permanent. Perhaps they change about occasionally without mutating. If so, then one type would disappear and the other would be formed anew somewhat in the same way as happens more regularly in the case of the two-type mating strains, but unlike what one would expect of genes or kappa-type bodies.

It has been assumed that the antigenic types (*A, B, C, D*) in *P. aurelia* are due to kappa-type bodies (Sonneborn and Le Suer, 1948). It is true that an antigenic type persists through many conjugations and autogamies and that it remains within its own line through conjugation, just as the kappa bodies do. However, it is again possible that here, as with the mating types in the *B* races, an autocatalyst of a more persistent type is involved and that it requires some unusual environmental condition (such as an antibody) to get rid of it, but that it can again be restored under genic influence in combi-

nation with the proper environment. In order to prove that the antigenic types are really due to kappa-type bodies, it would have to be shown that a line can lose a given type of antigen by speeding up its division rate just as in the case of kappa, itself. It would further have to be shown that a line could not regain its antigenic type once it had completely lost it. Moreover, it would be desirable to know whether the antigenic types, upon being stained, are found to contain visible bodies similar in size and number to those described by Preer for the killer race. Similar considerations would apply in connection with the mating types in the *B* races.[1]

Traits that are artificially induced in Paramecium by environmental agents sometimes persist through many sexual cycles. These are the *Dauermodificationen* of Jollos. They eventually disappear under normal conditions, and so are not due to any permanent genic change. They might, however, be due to non-genic autocatalysts, similar in principle to those that control mating type in the two-type strains, but again of a more persistent type. They could hardly be due to the loss of kappa-type bodies (as when killers become sensitives by the loss of kappa), for their return to normal would imply a restoration of the kappa-type bodies, and we saw that kappa cannot be made to reappear in a line once it has been lost (unless new kappa is introduced into the line by delayed conjugation).

It is conceivable that there are kappa-type bodies for other traits in addition to the killer trait, but if such bodies are symbionts one might often expect competition between them, so that eventually only one type would tend to persist in a given line. As a matter of fact, the number of different kinds of kappa-type bodies in a given

[1] Since this article was submitted for publication (Oct. 27, 1948) it has been reported that the antigenic types can, after being lost, be formed *de novo* under genic influence (Sonneborn, 1949), and it is therefore evident that they are due to non-genic autocatalysts, an interpretation which Sonneborn now accepts in effect, though he regards the bodies in question as a kind of gene.

Paramecium does appear to be very limited, since kappa itself (including its mutant forms) is the only well-established body of its kind.

A seemingly strange situation in the genetics of Paramecium is the fact that in the *A* races (those without kappa-type bodies) all known cases of inheritance are Mendelian, but in the *B* races (those with kappa-type bodies) all known cases are cytoplasmic. This has been interpreted as indicating a fundamental difference in the nature of the genic material and in the mechanism of inheritance in the *A* and *B* races. It would indeed be strange if the usual Mendelian mechanism had been displaced in the *B* races by another mechanism (of kappa type), or if it had been relegated to a position of secondary importance and rendered impotent as an hereditary mechanism, particularly since the *B* races are very closely related to the *A* races, in which the Mendelian mechanism still holds good; and further since the *B* races possess a nuclear structure that is visibly identical with the one possessed by the *A* races and that serves in them (the *A* races) as the material basis of Mendelian inheritance.

In this connection it should be noted that there are not many known cases of simple clear-cut Mendelian inheritance even in the *A* races of *P. aurelia.* This is due partly to the fact that many of the differences between the various lines of Paramecium (made pure by autogamy) are of a quantitative nature and are probably due to multiple factors. Crosses between such lines would therefore not give evidence of simple Mendelian inheritance. By contrast, kappa would show clear-cut cytoplasmic inheritance. But actually the vast majority of traits would still be gene-controlled and the usual mechanism of inheritance would still operate.

If the above account is correct, then there is nothing unusual about the genic system of Paramecium proper. The reduction divisions take place as usual for other organisms before gamete formation (during conjuga-

tion, autogamy, or cytogamy), and they are accompanied by Mendelian segregation. There is no conclusive evidence for segregation at any other time. Neither is there conclusive evidence for plasmagenes, if by this term we mean genes within the cytoplasm belonging to the genetic system of Paramecium proper and not to a symbiont.

In conclusion, then, the ciliates conform with other organisms in regard to the fundamental processes of heredity and development. Nevertheless, the findings of Sonneborn and his co-workers rank among the important discoveries in modern genetics, in that they reveal the amazing extent to which symbionts might become enmeshed in the heredity and development of their host, and in that they call attention to experimental findings which can be interpreted as confirmatory evidence for the theory that the inheritance of differentiation in asexual reproduction is due to autocatalysts of a non-genic nature.

Summary

Accumulating evidence supports the theory that kappa is a symbiont, rather than a plasmagene. The inheritance of differentiation in Paramecium is not due either to kappa-type bodies or to plasmagenes, but more likely to autocatalysts of a non-genic nature. This is indicated by the fact that in the two-type mating strains the material basis of differentiation (into a plus or a minus type) is inherited during asexual reproduction but disappears at conjugation, and is then formed *de novo* presumably under the influence of nuclear genes in conjunction with the local environment. It is possible that the same explanation applies to mating type in the *B* races, as well as to the four antigenic types (*A, B, C, D*), but that in these cases the autocatalyst would be of a more persistent type. It would, however, still be non-genic, provided it could arise *de novo* after having disappeared.

It is extremely improbable that a totally new mechanism of heredity, cytoplasmic in nature, has displaced

the Mendelian mechanism in the *B* races, particularly in view of the similarity of their chromosomal behavior to that of the *A* races, in which the usual mechanism of Mendelian inheritance is found.

LITERATURE CITED

Altenburg, E.
1946. "The Symbiont Theory in Explanation of the Apparent Cytoplasmic Inheritance in Paramecium," AM. NAT., 80: 661–662.

Jennings, H. S.
1938. "Sex Reaction Types and Their Interrelations in *Paramecium bursaria*." *I. Proc. Nat. Acad. Sci.*, 24: 112–117.

Muller, H. J.
1929. "The Gene as the Basis of Life," *Proc. 11th International Congr. Plant Sci.*, (Ithaca), 1: 897–921.

Preer, J. R.
1946. "Some Properties of a Genetic Cytoplasmic Factor in *Paramecium*," *Proc. Nat. Acad. Sci.*, 36: 247–253.
1948*a*. "The Killer Cytoplasmic Factor kappa," AM. NAT., 82: 35–42.
1948*b*. "A Study of Some Properties of the Cytoplasmic Factor 'Kappa' in *Paramecium aurelia*, Variety 2," *Genetics*, 33: 349–404.

Sonneborn, T. M.
1949. "Beyond the Gene," *Am. Scientist*, 37: 33–59.

Sonneborn, T. M., and A. Le Suer
1948. "Antigenic Characters in *Paramecium aurelia* (Variety 4): Determination, Inheritance and Induced Mutation," AM. NAT., 82: 69–78.

INHERITANCE IN CROSSES OF JERSEY AND HOLSTEIN-FRIESIAN WITH ABERDEEN-ANGUS CATTLE. III. GROWTH AND BODY TYPE, MILK YIELD AND BUTTERFAT PERCENTAGE[1]

LEON J. COLE[2] AND IVAR JOHANSSON[3]
UNIVERSITY OF WISCONSIN

INTRODUCTION

WHEN the Wisconsin crossbreeding experiment was planned and started in 1912 the main object was to study the inheritance of such economically important quantitative characters as rate of growth, beef qualities, milk yield and butterfat percentage. The calves were weighed at birth and weekly thereafter. A number of different measurements were taken of each individual at one and four weeks of age, and then at four-week intervals until the age of one and a half years. Later on measurements were taken at 21 and 24 months of age and thereafter annually. The animals were judged by a member of the Animal Husbandry Department using the score card of this institution. Slaughter data on the weight of hide and internal organs, as well as of the carcass and cuts, were obtained in all cases where possible.

The milk of each lactating cow was weighed at each milking and a sample was taken for determination of the butterfat content. The Babcock test was made once a week on a preserved, composite sample. In the early part of the experiment a three-day composite sample was taken of the milk from each cow once in every four-week period and analysed for total solids, butterfat, total protein, casein, milk sugar and ash. Also, the iodine (Hübl) and Reichert-Meissl numbers of the butterfat were de-

[1] Paper from the Department of Genetics, University of Wisconsin, No. 399.

[2] Deceased February 17, 1948.

[3] Professor of Animal Breeding at the Agricultural College of Sweden.

termined. The chemical analyses were discontinued in 1922, however, due to the cost involved.

The cows in the crossbreeding herd were stall-fed practically the whole year on hay, silage and a suitable concentrate mixture. As a rule only dry cows, heifers, and in some cases steers, were pastured. Silage and grain were fed according to live weight and milk yield of the animal, and hay was provided *ad libitum*. Care was taken that the grain ration contained several different feeds and had a suitable protein balance. The aim was to keep the environment as constant as possible from year to year and for all animals in the herd, maintaining good farm conditions of feeding and management.

At attempt was made to keep all females of the F_1 and F_2 generations until they had completed at least two lactations. F_1 and F_2 bulls which were not needed for breeding were castrated before four months of age and raised as steers. During the early part of the experiment the steers were finished and slaughtered at about 18 months of age but during the last part, somewhat earlier. When the experiment was discontinued in the spring of 1933 all animals were slaughtered regardless of age.

At the time the crossbreeding herd was disposed of it was fully realized that the number of animals in the F_1 and F_2 generations was far too small to make possible an analysis of the inheritance of the quantitative characters for which records were taken, but it was felt that the additional information that could be obtained by continuing the experiment would not be proportionate to the cost involved. Although no definite conclusions can be drawn on the basis of the rather meager data at hand, a summary of the results will be presented, because they are of interest in relation to similar data from other crossbreeding experiments. No attempt will be made in this connection to review the literature on crossbreeding or on the inheritance of quantitative characters in cattle. All data on the experimental animals are available at the Department of Genetics, University of Wisconsin.

Growth and Type of the Animals

To give an indication of the size and type of the experimental animals average figures on live weight, body length and wither height of mature cows are presented in table 1, together with the dimension-weight index,[1] calculated according to Yapp (1924), the total score when the cows were judged for "dairy qualities," and the score for udder development. The scores are stated in per cent of the maximum score possible.

TABLE 1

Average Live Weight, Measurements and Scores of Mature Cows

	Number of cows	Live weight lbs.	Body length cm.	Wither height cm.	Dimension-weight index	Total score	Score for udder
Aberdeen-Angus	7	1265	151	122	3.76	71.0	62.3
Jersey	2	1015	144	126	4.76	89.0	85.0
Holstein-Friesian	6	1293	161	132	4.61	82.3	74.9
F_1 Jersey × Angus	4	986	148	120	4.57	83.4	82.2
F_2 Jersey × Angus	4	1045	144	120	4.30	78.6	71.1
F_1 Holstein × Angus	17	1322	159	130	4.30	74.6	67.9
F_2 Holstein × Angus	14	1295	155	128	4.17	73.5	64.0

The score for udder development is surprisingly low for the foundation Holstein cows. The milk records (Table 4) show also a rather low yield for these cows, compared to what would be expected from purebred cows of the Holstein breed. Unfortunately high class animals could not be procured as foundation stock, due to lack of funds. The variation in type and milking ability of the foundation cows was rather pronounced. Data on milk yield and butterfat percentage of the individual foundation Angus cows, as compared to the Jersey and Holstein herd-mates, are presented in an earlier publication (Cole and Johansson, 1933). The variation in type of the Angus cows may be seen from figs. 1 and 2. The Angus cow 3A (Fig. 2) was of good beef but a very poor milker, whereas the cow 35A (Fig. 1) was more of a dual purpose type and a fairly good milker.

[1] The dimension-weight index is the ratio between the calculated volume of the animal and a rectangular prism having the squared wither height of the animal as base and the body length as height. The index is comparatively low for the blocky type and high for the rangy type of animals.

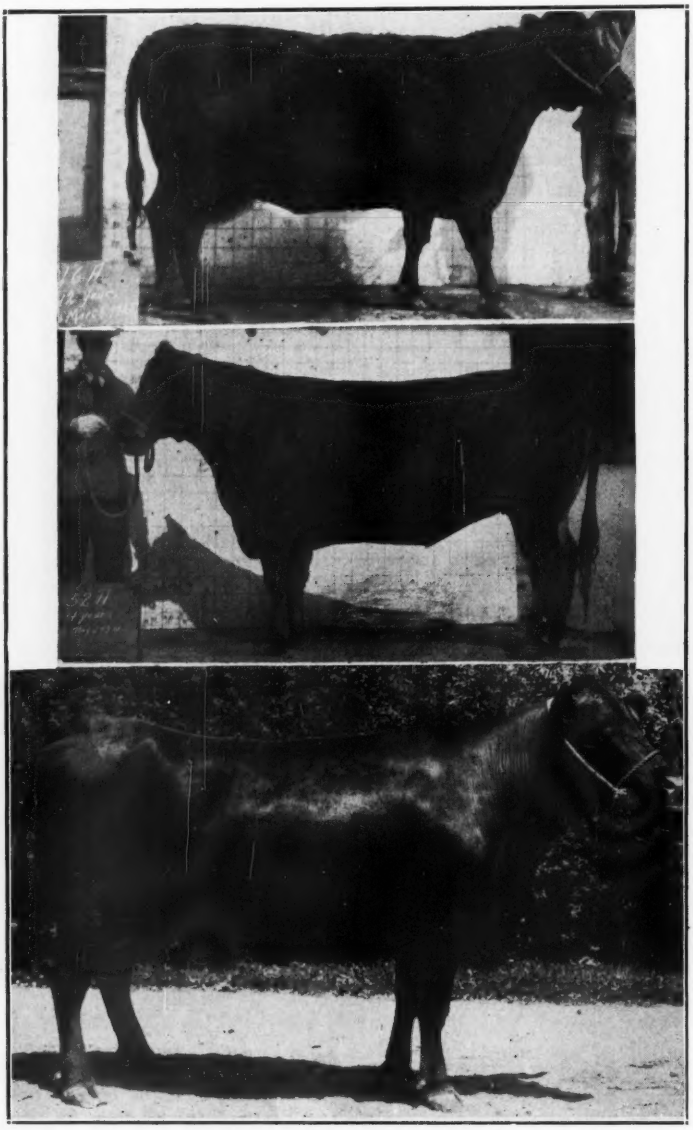

FIG. 1. Three foundation Angus cows in the Holstein × Angus cross: 16A, 32A and 35A.

The number of animals in the Jersey × Angus cross is obviously far too small for a statistical treatment of the data. Fig. 2 gives an idea of the difference in type of the foundation Angus cows and their daughters, sired by a Jersey bull. On the whole the type of the F_1 animals was intermediate to that of the parental breeds.

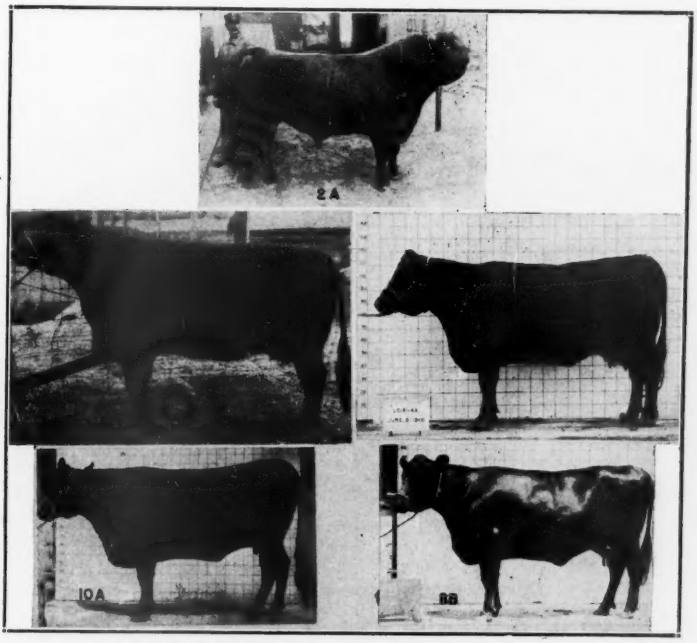

FIG. 2. Foundation and F_1 animals from the Jersey × Angus cross: Jersey bull 2A, Angus cows 3A and 4A and their F_1 daughters 10A and 8B, sired by 2A.

An attempt was made to analyze the data from the Holstein × Angus cross. Only data on birth weight and heart girth of crossbred animals will be presented in this paper. No statistically significant differences between F_1 animals from reciprocal crosses, or between the standard deviations of the F_1 and F_2 generations, were found in regard to these characters, and therefore no analysis of the other measurements was made.

For the F_1 and F_2 cows of the Holstein × Angus cross coefficients were calculated for the correlation between milk production and body weight (–0.095), dimension-weight index (0.142), total score (0.030) and score for udder development (0.125). None of the coefficients is statistically significant.

The average gestation period and the birth weight of the calves from the Holstein × Angus cross are presented in table 2. No comparable figures for birth weight of the

TABLE 2

LENGTH OF GESTATION PERIOD AND BIRTH WEIGHT OF CALVES IN THE HOLSTEIN X ANGUS CROSS

Generation	Number of calves	Bull calves				Number of calves	Heifer calves			
		Length of gestation, days		Birth weight lbs.			Length of gestation, days		Birth weight lbs.	
		Ave.	S.D.	Ave.	S.D.		Ave.	S.D.	Ave.	S.D.
F_1 Angus ♂ × Holstein ♀	6	281.2	3.97	84.5	10.74	11	279.2	6.68	75.9	10.66
Holstein ♂ × Angus ♀	7	277.6	6.90	82.0	11.12	11	276.4	5.57	69.5	11.98
F_1 average	13	279.2	5.76[1]	83.2	10.95[1]	22	277.8	6.15[1]	72.7	11.38[1]
F_2	19	278.6	5.64	79.3	9.71	23	278.0	6.02	71.0	11.29

[1] Standard deviation within groups from the reciprocal crosses.

foundation animals are available. The gestation periods for F_1 calves agree fairly well with corresponding averages from matings within the Holstein and Angus breeds, published by Livesay and Bee (1945). The differences in birth weight between male F_1 calves of the Angus ♂ × Holstein ♀ and the Holstein ♂ × Angus ♀ cross is 2.5 ± 6.07 lbs. and the corresponding difference for the female calves is 6.4 ± 4.83. The differences obtained on these small numbers are not significant. When the sum of squares within groups are pooled, thus eliminating differences between reciprocal crosses and the sex groups, the standard deviation in birth weight for the F_1 generation is 11.20 and for F_2, 10.61. Thus there is no evidence of increased variation in F_2 in regard to the birth weight of the calves.

Fig. 3 shows the average growth in heart girth of female F_1 animals from the reciprocal crosses. The heart girth of F_1 cows from the Holstein ♂ × Angus ♀ cross (9 animals) is consistently higher than the re-

ciprocal (10 animals) except at birth; the difference increases until about 12 months of age when it reaches statistical significance. This is quite remarkable be-

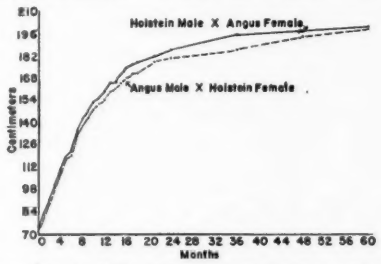

FIG. 3. Average heart girth of F_1 cows from the reciprocal crosses Holstein × Angus and Angus × Holstein.

cause the Angus cows were smaller in size (heart girth) than the Holstein cows (Fig. 4) and therefore rather the reverse result would have been expected. The variation within the two groups was not significantly different.

Fig. 4 presents a comparison between the growth in

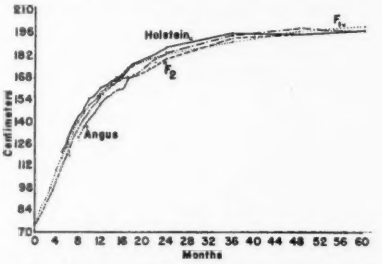

FIG. 4. Average heart girth of foundation Holstein and Angus cows and of the F_1 and F_2 generation females.

heart girth of foundation cows and corresponding averages for females of the F_1 and F_2 generations. Data were available for three Angus cows only from eight months of age and for four Holstein cows from five months. For 12 F_1 and 16 F_2 females data were available from birth to 48 months of age. The growth curve for the F_1 cows comes closer to the Holsteins than to the Angus, but it must be remembered that the averages are calculated on small numbers. The curve

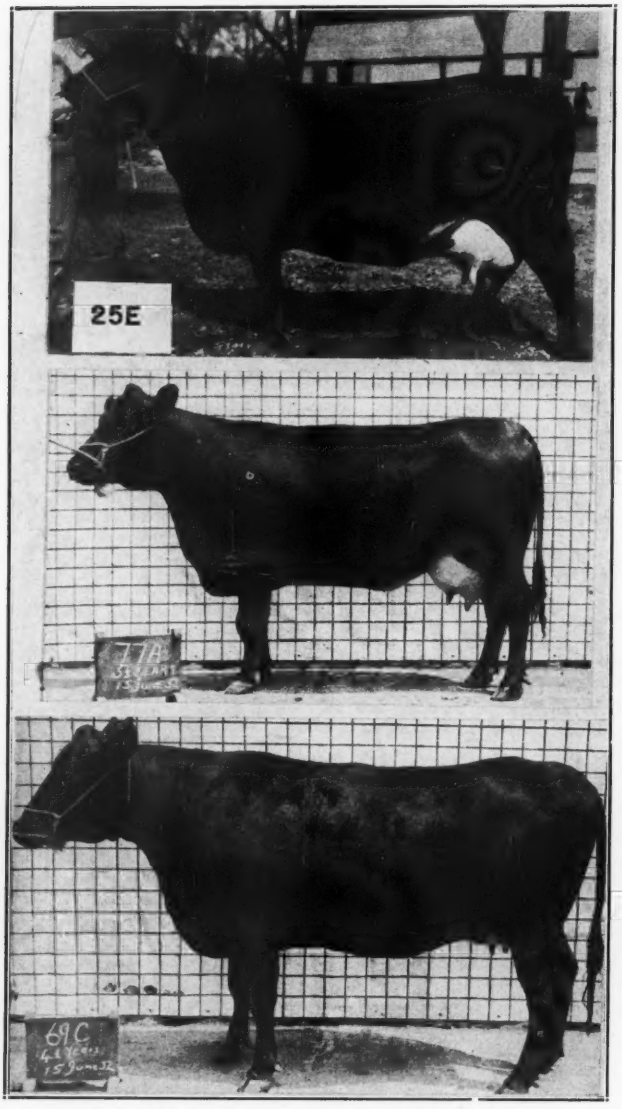

FIG. 5. F_1 cows of the Holstein × Angus cross: 69C, 77A and 25E. All three intermediate in type to the parental breeds.

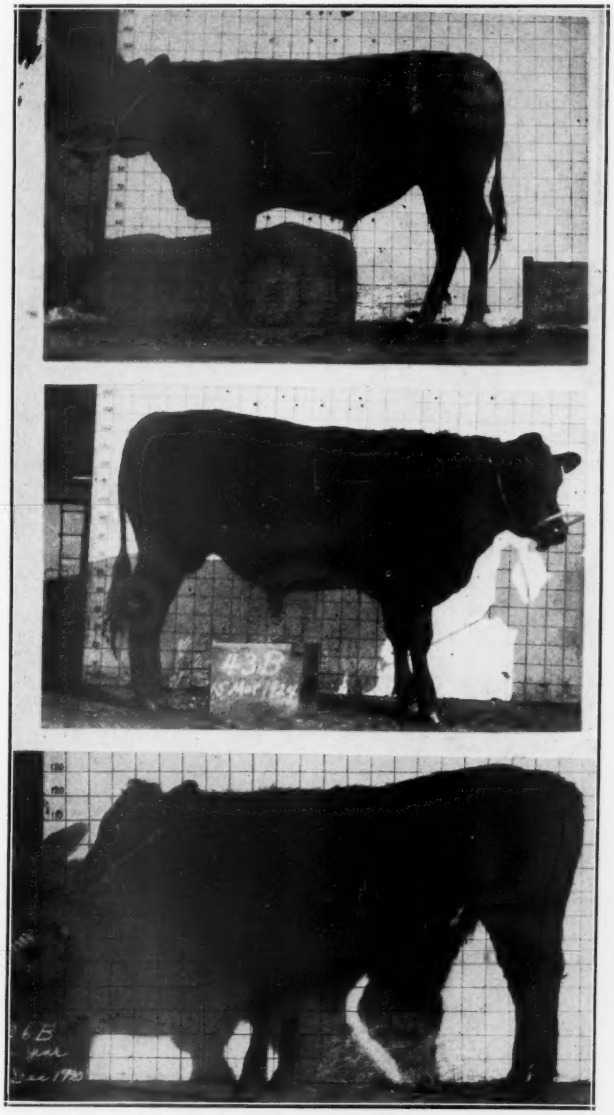

FIG. 6. Three F_1 steers from the Holstein-Angus cross: 26B, 43B and 43A, all 12–14 months of age.

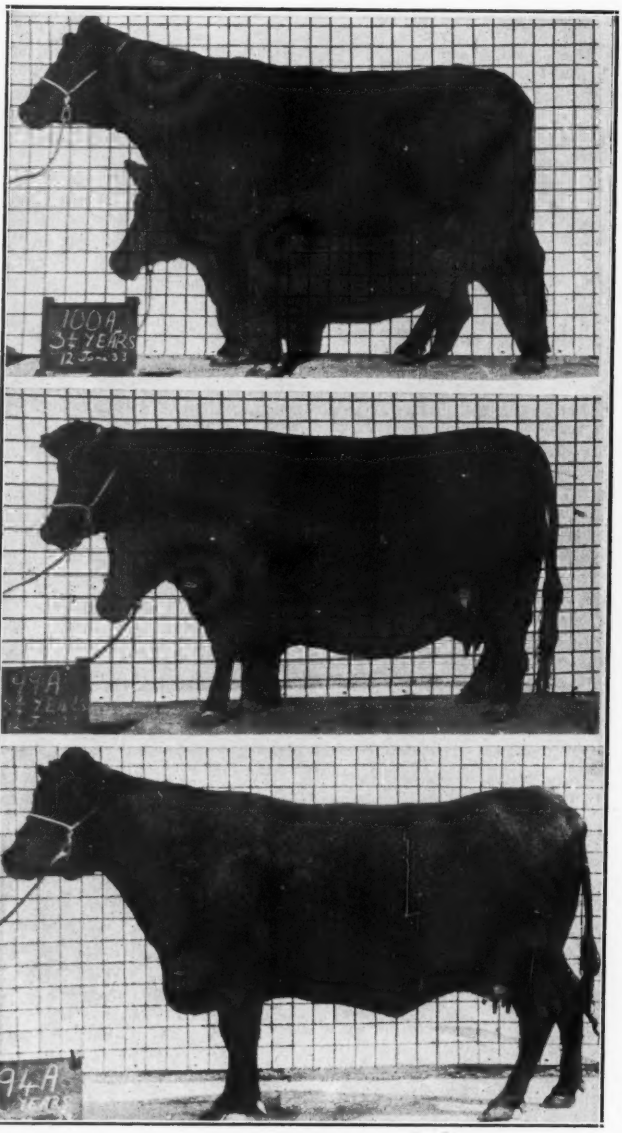

FIG. 7. Three F_2 cows from the Holstein × Angus cross: 94A, 99A and 100A. The cow 94A tends towards the Angus type and 100A towards the Holstein type, whereas 99A is intermediate.

for the F_2 generation is consistently lower than that for the F_1 animals. There is no significant difference in the standard deviation of the F_1 and F_2 groups.

Figs. 5–7 give a general idea of the type of F_1 and F_2 animals from the Holstein × Angus cross. Fig. 5 shows three F_1 cows and Fig. 6, three F_1 steers. In Fig. 7 cow 94A represents the "beefy" and 100A the dairy type, whereas 99A is intermediate. An examination of the body measurements and scores for type does not reveal, however, any greater variation in the F_2 than in the F_1 generation. Therefore there is no real evidence for a segregation for type in the F_2 generation.

All that can be said on the basis of the analysis of the data on growth and beef qualities is that the F_1 animals are more or less clearly intermediate to the parental breeds, and that no evidence of increased variation in F_2 could be established. There are some differences between the F_1 animals from reciprocal crosses, but the statistical evidence on this point is rather slight.

Milk Yield and Butterfat Percentage

In the Jersey × Angus cross there are only four F_1 and four F_2 cows with lactations after normal calving; in the Holstein × Angus cross the corresponding numbers are 17 and 14.

Due to a rather great variation in the length of calving interval, it was decided to use the milk yield and butterfat percentage in the first 180 days after calving as a measure of the performance of the cow. Also, the total lactation yield was calculated but will not be presented here. The criticism can be made that taking the yield in the 180 days after calving as standard tends to favor the Angus breed more than the Jerseys or Holsteins, due to difference in persistency of yield between the beef and dairy breeds as shown in fig. 9. The error involved was considered less, however, than if using the total lactation yield, or the yield in 305 days, omitting correction for variation in length of the calving interval. The milk yield in 180 days was corrected to the same energy basis

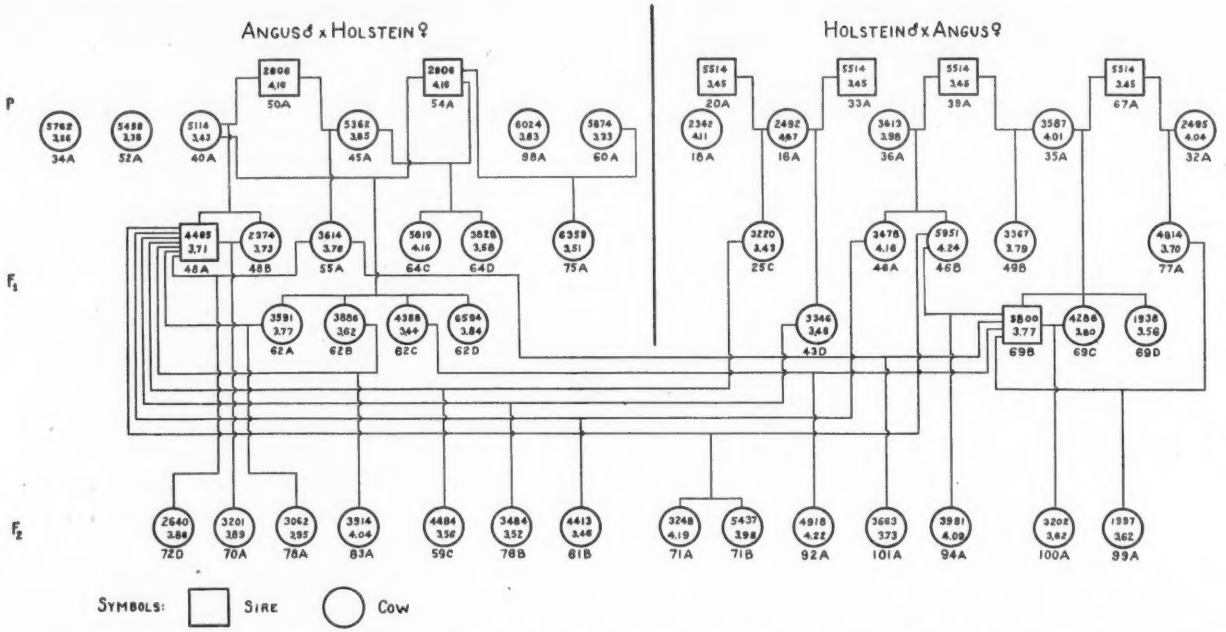

FIG. 8. Chart showing the mating combinations in the Holstein × Angus cross for the production of F_1 and F_2 cows, each cow with at least one normal lactation. For the cows, figures are stated which show the average milk yield and butterfat percentage in 180 days of their first three lactation periods. The figures for Angus and Holstein bulls show the average production of the foundation cows of the sire's own breed, and the figures for F_1 cows show the average production of all F_1 cows from the same cross as the bull.

according to Gaines and Davidson (1923), i.e., to milk with 4 per cent butterfat. For each cow with complete

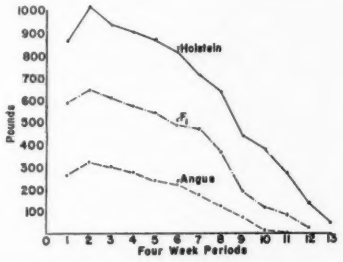

FIG. 9. Average lactation curves for five Holstein, four Angus and seven F_1 cows from the Holstein × Angus cross.

records for the first three lactations the average yield of these lactations was taken as a measure of the cow's producing ability. Where the number of lactations was less than three, the yield was age-corrected on the basis of the yield in the first three lactations of 10 cows, representing all the breeds and crosses in the experiment.

TABLE 3

AVERAGE MILK YIELD AND BUTTERFAT PERCENTAGE IN THE JERSEY × ANGUS CROSS

	Average yield in 180 days of the first three lactations	
	Fat corrected milk, lbs.	Butterfat percentage
4 Angus cows	2030 ± 374	4.32 ± 0.12
2 Jersey cows	4528 ± 949	4.82 ± 0.18
4 F_1 cows	3147 ± 348	4.37 ± 0.26
4 F_2 cows	3629 ± 427	4.26 ± 0.20

The average yield of the cows in the Jersey × Angus cross is shown in table 3.

The averages of the F_1 generation fall below those of the two parental breeds in milk and butterfat percentage.

The detailed results of the Holstein × Angus cross are shown on the accompanying chart (Fig. 8). In this chart "potential yield" figures are given for the sires. The figures for the Aberdeen-Angus sires are simply the average milk yield and butterfat percentage of all the foundation Angus cows in the crossbreeding herd, and the figures for the Holstein-Friesian sires are the averages of all the foundation Holstein cows. The figures

for the F_1 bull 48A are averages for all the F_1 cows from the Angus ♂ × Holstein ♀ cross, and for the F_1 bull 69B the figures are averages for all the F_1 cows from the same cross as the bull (Holstein ♂ × Angus ♀).

A summary of the results is presented in table 4.

TABLE 4

AVERAGE MILK YIELD AND BUTTERFAT PERCENTAGE IN THE HOLSTEIN × ANGUS CROSS

	Average yield in 180 days of the first three lactations			
	Fat corrected milk, lbs.		Butterfat percentage	
	Mean	S.D.	Mean	S.D.
5 Angus cows	2906 ± 284	637	4.16 ± 0.13	0.288
6 Holstein cows	5599 ± 141	345	3.51 ± 0.11	0.262
F_1:				
9 Angus ♂ × Holstein ♀	4495 ± 480	1438	3.71 ± 0.07	0.215
8 Holstein ♂ × Angus ♀	3800 ± 426	1207	3.77 ± 0.11	0.298
Ave. for 17 F_1	4168 ± 325	1341	3.73 ± 0.06	0.253
14 F_2 cows	3689 ± 246	917	3.84 ± 0.07	0.254

The differences between the parental breeds in milk yield (2693 ± 317) and butterfat percentage (0.65 ± 0.17) are highly significant. In the F_1 generation the average yield of fat corrected milk is somewhat higher for cows from Holstein dams and Angus sires than in the reciprocal cross, but the difference is not significant. In butterfat percentage these two groups are approximately equal. There is no indication that the sire has had a different effect than the dam on the yield and composition of milk of the daughters. Most F_1 cows are intermediate to their parents in milk yield and butterfat percentage, but there is considerable variation. There is no evidence in support of the hypothesis that high milk yield and low butterfat percentage show a certain degree of dominance. Likewise there is no statistically significant difference in variation between the F_1 and F_2 generations.

If one or more of the major genes for milk yield or butterfat percentage were sex-linked as suggested by Smith and Robison (1931), a difference would be expected between cows that had an Angus and those that had a Holstein cow as paternal grandam. Among the 14 F_1 cows with milk records there are nine with an

Angus and five with a Holstein grandam. The averages for these two groups are as follows:

	Average yield in 180 days	
	Milk, lbs.	*Butterfat percentage*
5 F_2 cows with paternal Angus grandam	3765 ± 293	3.83 ± 0.09
9 F_2 cows with paternal Holstein grandam	3552 ± 479	3.86 ± 0.11

The differences between the two groups are not statistically significant.

Fig. 9 shows the average yield of milk in four-week periods of the first lactation for four Angus, five Holstein and seven F_1 cows from the Holstein × Angus cross, none of these cows having longer lactation period than 365 days. The Holstein cows differ from the Angus more in the level of yield than in the shape of the curves. The Angus cows have, however, a comparatively short lactation and a long dry period.

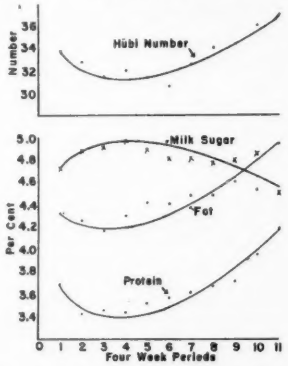

FIG. 10. The variation in the composition of the milk during the lactation period: protein, fat, milk sugar and the iodine (Hübl) number of the butterfat. Average for 16 lactations.

For 16 lactations of 10 different cows in the Jersey × Angus cross, complete data are available for the chemical analyses of the milk every fourth week throughout the lactation. Although the data are too limited for an analysis of the genetic variation between individuals, the averages for all lactations may be of interest, because the cows were fed the same ration throughout the whole lactation. Fig. 10 shows the systematic changes

in the milk sugar, fat and protein content of the milk, and in the iodine number of the butterfat. The fat and protein content of the milk show the same general trend during the lactation, the percentages being lowest at the time of maximum daily yield and highest towards the end of the lactation. The trend in the iodine number is similar to that of the butterfat percentage.

Summary

Data are presented on the birth weight, heart girth, milk yield and butterfat percentage of animals in the Wisconsin cattle crossbreeding experiment. In the Holstein $\times$ Angus cross F_1 daughters of Angus dams showed a slightly higher heart girth than F_1 daughters of Holstein dams. In milk yield and butterfat percentage no significant difference between cows from the reciprocal crosses was found, and the variation was not greater in the F_2 than in the F_1 generation. Most F_1 animals were intermediate to the parental breeds in all the characters studied.

LITERATURE CITED

Cole, L. J. and Johansson, Ivar
1933. *J. Dairy Sci.* 16: 565–580.

Gaines, W. L. and Davidson, F. A.
1923. *Univ. of Ill. Agric. Exp. Sta. Bull.* 245: 575–621.

Livesay, E. A. and Bee, U. G.
1945. *J. Animal Sci.* 4: 13–14.

Smith, A. D. B. and Robison, O. J.
1931. *Proc. Int. Dairy Congress,* English Ed., Section 1: 127–140. Copenhagen.

Yapp, W. W.
1924. *Proc. Amer. Soc. Animal Prod.* 1923: 50–56.

CATCHING FISHES WITH THE HAND[1]

I. IN THE TWO AMERICAS: 1699–1942

DR. E. W. GUDGER
AMERICAN MUSEUM OF NATURAL HISTORY

INTRODUCTION

SOME time ago a letter came to the Museum asking the settling of a controversy as to whether it is possible, by putting the hand gently into the water on the shady side of a brook, to have a fish "cuddle" in it and thus be easily caught.

My interest in unusual fishing methods being well known, the query was referred to me. By good fortune I had first hand knowledge of this practice when a boy. Furthermore, I have a large amount of data bearing on this fishing the world around. Hence I was able to answer the correspondent's query in the affirmative.

It seems worth-while to prepare a series of articles bringing together all the available accounts of this practice in widely separated parts of the world, setting them down in an orderly review as a contribution to the history of this most primitive fishing method. This seems all the more worth-while, since no one, not even an ethnologist, so far as I can find, has ever attempted this task.

Some of the data at hand reach far into the past and establish this fishing habit as more or less widespread over three continents and in certain regions of smaller size. It is natural that, in a series of such articles one should begin with his home continent. But, in doing so and in order to get off to a good start, it will be better for once to reverse the chronological order of presentation, and for North America to begin with some present-day accounts, and particularly to describe the exploits

[1] By fishing with the hand, just that is meant—the use of the human hand exclusively, without the aid of any auxiliary instrument. In the one exceptional case herein, special note is made.

of a boyhood playmate and some feeble ones of the present writer.

Fishing with the Hand in North America

So far as I have found, this fishing method does not seem to have been practiced by the Indians of North America north of the Rio Grande nor by the Toltecs and Aztecs south of that river. However, it was and is practiced in the U.S. by the descendants of English-speaking settlers. And we shall find it among the Indians in the southernmost part of our continent.

Hand-fishing in the United States

In the southern Appalachian Mountains, my home town, Waynesville, is situated on the high plateau of Western North Carolina. Seventy years ago all the streams in that whole section abounded in fishes, speckled or brook trout especially. About a mile from my home, my playmate, John Norwood, lived almost on the bank of a lovely brook filled with fishes. I was there often, and in that brook he and I, literally "barefoot boys," used to wade and "grabble" (as we termed it) for fishes. These took refuge under stones, roots, stumps, submerged logs and overhanging banks. Here we found the fishes and brought them out with our hands. I, from infrequent practice, was not very successful, but John, who was in the brook almost every day, was very skillful. And little did he know that he was doing what his English forebears had probably done hundreds of years before. Whether he "cuddled" or "tickled" the fishes I cannot recall after all these years. Nor can I remember whether other boys in our section caught fishes in this fashion, but it seems probable. And recently a man from a neighboring county wrote me that he has long known of this fishing but had never practiced it.

The practice seems to be more widespread in the United States than I had conjectured. At any rate in speaking of it casually to two friends here in the Mu-

seum, each said "Yes, I have caught fishes with my hands," and each has communicated an account in writing.

Mr. George Goodwin is English born and he practiced "tickling trout" in his boyhood homeland. Some 25 years ago he and a friend here were speaking of the gullible public which swallows as facts almost everything that is found on the printed page. His friend mentioned as a case in point "that ridiculous story about tickling trout." As to what then took place, Goodwin's personal communication is quoted:

To me this was no story of the fickle imagination, and to prove my point the two of us were soon on our way to a stream in the Orange Hills, New Jersey. At the very first stream we found an ideal spot where the swift-running water, draining from a deep pool, cut under the bank. Lying prone on the ground with my sleeve rolled up I reached down into the water, the back of my hand was on the gravel bottom and the tips of my fingers turned up. I felt cautiously under the bank and, moving gradually upstream so as to approach a trout from the rear if there was one, presently my finger tips ever so faintly touched the smooth underside of something that, though seemingly stationary, actually moved a fraction forward. Luck was with me, I had a trout almost within my grasp. Slowly and lightly my fingers caressed its smooth belly, working gradually forward. In five minutes my fingers had reached its gills, now my forefinger and thumb slowly crept up each side while my little finger continued to lightly massage the central belly line. Then, with a steady firm pressure, my thumb and forefinger locked into the gills and a ten-inch trout was lifted into full view of my astonished and now convinced friend.

Dr. H. E. Anthony has kindly communicated some boyhood experiences of fishing with the hand:

No one taught me, but as a boy nine years of age, I caught trout by hand in the Sierra Nevada of California. A clear-water brook contained a fair number of trout which frequented retreats under boulders and shelving rocks. I knew the location of the best of these and not

infrequently was able to catch fair-sized fish by bringing my hand across the opening of such a nook. When a fish was thus located it would attempt to push past the hand to open water. By careful manipulation of my fingers I could get a grip back of the gills and draw out the struggling fish. An attempt to pull out the trout with a grip on the body usually permitted the fish to slip through my fingers. It was necessary to slip the fingers to the gill-grip and this was best done by gentle movements rather than a rough gesture that made the fish desperate.

Professor H. H. Lane of the University of Kansas states in a personal communication: "My experiences in catching fish by hand are very few and occurred mostly when I was a boy in Indiana. In common with other boys, I would occasionally lie down on a flat rock or log and reach underneath it for sucker or shiner, which only occasionally was so penned in that it could not escape."

Turning now to another part of North America, to a region far removed in time and space from all those noted above, the two accounts will be set out in chronological order.

Diving and Fishing with the Hand in Panama

Lionel Wafer, a British traveler, went to Jamaica and in 1679 joined the buccaneers. In 1680 he crossed to the Isthmus of Panama with Captain Dampier and as a result of a quarrel was left in 1681 among the Indians with whom he lived until 1684. In 1699 he published the first good description of the Isthmus in English. Wafer was much interested in the natural history of the region and described many of the striking things new to him. His account of the fishes is good reading, and his description of a method of fishing in the rivers is especially interesting. He notes that when the Indians go along the banks of a river they are always looking for fishes and then he states that:

> . . . in the Hill-Country where the Streams are clear . . . they go along the Banks up the River looking nar-

rowly into the water to view the Fish. When they spy any to their Mind, they leap into the Water, and wade or swim up and down after them; and if the Fish, through Fright, betake themselves into the holes in the Banks, for Shelter, as they frequently do, the Indians feel them out with their Hands, and take them thence, as we do Chubs or Craw-fish in our [English] Rivers. By Night they bring with them Torches of Light-wood, and with these they spy out the Fish and so jump in, and pursue them into their Holes.

Dr. C. M. Breder, Jr., head of the Department of Fishes in this Museum, some 225 years later spent some months collecting and studying the fishes of the Rio Chucunaque drainage on the Pacific side of the Isthmus. In the section of his paper (1924) dealing with the fishing methods observed, he has an interesting account of the methods used by the Chocoi Indians in certain side streams of the lower main river. Of the one of interest to us, he writes that even without spears:

> . . . they are still able to catch fish with comparative ease by resorting to the simple expedient of diving overboard and catching them with their hands. The loricates [catfishes] are usually the target for such activity, and the divers invariably bring the fish up between their teeth, placing them there, head first, to enable them to have more freedom of hand-movement. Occasionally they even capture a cichlid in this manner. Just how they are able to do it I am at a loss to say, for all that could ever be seen was the diving overboard, followed by a great flurry in the water and an emergence the next moment with the fish.

Here it is seen that, in the same region, Breder two and a quarter centuries later corroborated Wafer's observations. Wafer does not explicitly state, as does Breder, that his Indians dived after their fishes but on reading his account I conclude that they did so. Wafer does not name the river of this fishing, but the account is in the chapter in which fishes of the north coast are described. Presumably he saw this fishing in the north-flowing rivers.

Since the above was written, my attention has been

called to a book, "The Lummi Indians of Northwest Washington" by B. J. Stern (1940). In this the statement is made (p. 51) that these Indians catch flounders by wading in the water and that they "hold the fish by stepping on them until they can pick them up and throw them into the canoes." This account is interesting in itself and also because it ties up with an account of feeling for fishes with the feet as recorded from China by Robert Fortune in 1847.

Hand-Fishing in South America

When one thinks of the continent of South America with its great river systems, one wonders if this fishing is not practiced there by the riverines. But in a wide reading of books of travel and exploration largely by and on these rivers, only three accounts of hand-fishing have been noted. However, in two articles on primitive fishing methods, two unlike accounts have been found of hand-fishing in the same region of South America.

In The Gran Chaco

In this ill-defined region, wherein Argentina, Bolivia and Paraguay meet, the Indians, according to Krause (1904, p. 154), practice hand-fishing and have adopted an interesting device to enable them to secure their slippery prey. Thus: "The Lengua Indians, to prevent the escape of the fish when fishing with the hand, wear around the hand a band set with small animal vertebrae. The projections of the vertebrae hold the slippery fish easily." This is indeed a very clever and efficient device, since with the bare hand it is impossible to hold the fish unless the grip is in the gill-region.

Years later a slightly different hand-fishing in this area was described by L. C. Beadle (1929) in an article on the many ingenious fishing methods used by the Lengua Indians in the Gran Chaco. Of that method in which our interest centers, Beadle writes as follows:

The simplest and least skilful method employed on

these occasions [the oncoming of droughts and the concentration of water and fishes in pools] is known in the Lengua language as 'Pakningwukme' or 'feeling for the fish.' A party of men strip themselves and surround a clump of rushes, squat down and merely feel among the stems with their hands for fish hiding there, and all the time a great deal of chattering and talking goes on. When each fish is caught it is killed in a somewhat barbarous fashion. The head is put into the man's mouth and is bitten through[2]. The fish is then either thrown ashore or placed in a string bag, which is often carried for fish and game. The smaller fish, such as Callichthys [a catfish], are usually killed in this way.

In Lake Juin, Central Peru

This lake, whose native name is Chinchaycocha, is near Cerro de Pasco on the high plateau of Central Peru. Eigenmann and Allen (1942, pp. 1–2) report that they had great difficulty in collecting fishes in this shallow, mud-bottomed lake, the margin of which was overgrown with a closely-matted jungle of aquatic plants. Beyond this, numerous fishes could be seen, but could not be reached with hook and line, with nets, or dynamite. In this dilemma, "a Peruvian came to our rescue. With a hardihood inherent in the dwellers of the bleak pampas, he stepped into the water to his thighs, supporting his weight on the rhizomes and roots of plants. Here he searched among the stems for the fishes, and found them"—as his ancestors probably had done in this very lake in the days of the Incas.

In a personal letter, Prof. Allen makes the matter even clearer: "The lake was almost enclosed on every side by shore and emergent plants. A false bottom or mattress of roots extended considerable distances into the lake off shore, forming a tangled mass sufficient to bear the weight of the Indian alluded to . . . The Orestias (Poecilid) fishes were occupying holes among the roots out of which the Indian picked them. The natives

[2] Here note Breder's account of a somewhat similar habit of the Indians of Panama.

were so familiar with this method of collecting that there is no doubt of its being employed by them as the standard fishing procedure for that locality." Here then is established a second locality for fishing with the hand in South America.

This concludes the story of hand-fishing in the two Americas. Other articles in this series are in preparation and will appear in sequence as publication can be had.

LITERATURE CITED

Beadle, L. C.
1929 "Fish and Fishermen of the Paraguayan Chaco." *Salmon and Trout Mag.*, London. (Fishing with the Hand, pp. 237–238).

Breder, C. M.
1927 "The Fishes of the Rio Chucunaque Drainage, Eastern Panama." *Bull. Amer. Mus. Nat. Hist.*, Vol. 57, (Indians catching fishes with their hands, p. 153).

Eigenmann, C. H. and W. R. Allen
1942 Fishes of Western South America. Lexington, K. (Fishing with the Hand in Lake Juin, pp. 1–2).

Krause, Edward
1904 Vorgeschichtliche Fischereigerate und neuere Vergleichstucke, Verglichende Studie als Beitrag Geschichte des Fischereiwesens. *Zeitschr. Fisch, Hilfswissen*, Berlin, Vol. 11, 108 p., 4 figs.

Wafer, Lionel
1699. New Voyage and Description of the Isthmus of Panama, etc. London. (Indians catch fishes with the hands, p. 129). Hakluyt Society ed., Oxford, 1934, p. 77.